

PATENT COOPERATION TREATY

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INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY


(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

REC'D 30 MAR 2005

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Applicant's or agent's file reference 21039WO		FOR FURTHER ACTION		See Form PCT/PEA/416
International application No. PCT/NL2004/000300		International filing date (day/month/year) 03.05.2004	Priority date (day/month/year) 01.05.2003	
International Patent Classification (IPC) or national classification and IPC C12N1/02, C12P1/00, C12M3/00, C12N5/00				
Applicant DSM IP ASSETS B.V. et al.				
<p>1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.</p> <p>2. This REPORT consists of a total of 6 sheets, including this cover sheet.</p> <p>3. This report is also accompanied by ANNEXES, comprising:</p> <p>a. <input type="checkbox"/> sent to the applicant and to the International Bureau) a total of sheets, as follows:</p> <p><input type="checkbox"/> sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).</p> <p><input type="checkbox"/> sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.</p> <p>b. <input type="checkbox"/> (sent to the International Bureau only) a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in computer readable form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).</p>				
<p>4. This report contains indications relating to the following items:</p> <p><input checked="" type="checkbox"/> Box No. I Basis of the opinion</p> <p><input type="checkbox"/> Box No. II Priority</p> <p><input type="checkbox"/> Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability</p> <p><input type="checkbox"/> Box No. IV Lack of unity of invention</p> <p><input checked="" type="checkbox"/> Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement</p> <p><input type="checkbox"/> Box No. VI Certain documents cited</p> <p><input type="checkbox"/> Box No. VII Certain defects in the international application</p> <p><input type="checkbox"/> Box No. VIII Certain observations on the international application</p>				
Date of submission of the demand 19.01.2005		Date of completion of this report 29.03.2005		
Name and mailing address of the international preliminary examining authority:  European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465		Authorized Officer Döpfer, K-P Telephone No. +49 89 2399-8547		



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Box No. I Basis of the report

1. With regard to the **language**, this report is based on the international application in the language in which it was filed, unless otherwise indicated under this item.
- ☐ This report is based on translations from the original language into the following language , which is the language of a translation furnished for the purposes of:
- ☐ international search (under Rules 12.3 and 23.1(b))
 - ☐ publication of the international application (under Rule 12.4)
 - ☐ international preliminary examination (under Rules 55.2 and/or 55.3)
2. With regard to the **elements*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report)*:

Description, Pages

1-6 as originally filed

Claims, Numbers

1-10 as originally filed

- ☐ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing
3. ☐ The amendments have resulted in the cancellation of:
- ☐ the description, pages
 - ☐ the claims, Nos.
 - ☐ the drawings, sheets/figs
 - ☐ the sequence listing (*specify*):
 - ☐ any table(s) related to sequence listing (*specify*):
4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).
- ☐ the description, pages
 - ☐ the claims, Nos.
 - ☐ the drawings, sheets/figs
 - ☐ the sequence listing (*specify*):
 - ☐ any table(s) related to sequence listing (*specify*):

* If item 4 applies, some or all of these sheets may be marked "superseded."

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Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes: Claims	1-10
	No: Claims	
Inventive step (IS)	Yes: Claims	1-10
	No: Claims	
Industrial applicability (IA)	Yes: Claims	1-10
	No: Claims	

2. Citations and explanations (Rule 70.7):

see separate sheet

Re Item I

Basis of the report

Re Item V

**Reasoned statement with regard to novelty, inventive step or industrial applicability;
citations and explanations supporting such statement**

1. Reference is made to the following documents:

- D1: US-A-5 378 612 (NAKASHIMA KAZUYUKI ET AL) 3 January 1995 (1995-01-03)
- D2: SAKAI KENTARO ET AL: "Use of nonionic surfactants for effective supply of phosphatidic acid in serum-free culture of Chinese hamster ovary cells." JOURNAL OF BIOSCIENCE AND BIOENGINEERING, vol. 92, no. 3, 2001, pages 256-261, XP002255475 ISSN: 1389-1723
- D3: US-A-5 372 943 (INLOW DUANE ET AL) 13 December 1994 (1994-12-13)
- D4: MICHİYUKI TOKASHIKI ET AL: "HIGH DENSITY CULTURE OF HYBRIDOMA CELLS USING A PERFUSION CULTURE VESSEL WITH AN EXTERNAL CENTRIFUGE" CYTOTECHNOLOGY, KLUWER ACADEMIC PUBLISHERS, DORDRECHT, NL, vol. 3, no. 3, 1 May 1990 (1990-05-01), pages 239-244, XP000117122 ISSN: 0920-9069
- D5: MICHİYUKI TOKASHIKI ET AL: "PERFUSION CULTURE APPARATUS FOR SUSPENDED MAMMALIAN CELLS" CYTOTECHNOLOGY, KLUWER ACADEMIC PUBLISHERS, DORDRECHT, NL, vol. 13, 1993, pages 149-159 cited in the application

2. Novelty and Inventive Step (Article 33(2)(3) PCT)

- 2.1 The present application addresses a process for the production of biological substances by perfusion culturing of suspended animal cells characterised by the presence of at least 0.001% of a polyoxyalkylene sorbitan fatty acid ester in the cell culture medium in order to prevent clogging of the filters when separating the cells from the supernatant.

2.2 None of the prior art documents cited discloses a process for the production of biological substances having the characteristic features (i) suspension culture of animal cells; (ii) comprising at least 0.001% of a polyoxyalkylene sorbitan fatty acid ester in the cell culture medium, and (iii) separation of the cells by filtration.

Claim 1 is therefore considered novel. The same applies to dependent claims 2-10 since they possess all technical features of the independent claim.

2.3 Document D5 has been identified as the closest prior art in that it has most features common with the present application. Furthermore addresses D5 suspension cultures of animal cells and the separation of the cells by filtration. The problem underlying the present application can be defined as to provide a method with improved filter performance. The problem with clogged filters is not only known from D5 but represents a problem which is commonly known to the skilled person in the art. The solution presented is the presence of at least 0.001% of a polyoxyalkylene sorbitan fatty acid ester in the cell culture medium.

The ant-clogging property of Tween 80®, which is a member of the chemical class of at least 0.001% of a polyoxyalkylene sorbitan fatty acid esters, is disclosed in D2 and D3 in respect to preventing the formation of aggregates and coagulates of lipids, i.e. the person skilled in the art gets a hint to the use of the Tweens® as anti-clogging agents. Nevertheless, it appears to be not obvious to the skilled person dealing with the technical problem (i.e. to prevent the clogging of filters during the separation of the cells from the liquid supernatant of a suspension cell culture) whether the above mentioned anti-aggregating property of Tweens® towards lipids would exhibit the same effect on the surface of filters. The prior art (see D4) discloses mechanical solutions, i.e. with centrifuges (other known measures are pulsed pressure, counter pressure). Even if taking into consideration that the anti-clogging property of e.g. Tweens® is an inherent one, the intended use for the presently claimed purpose has not been made public before or has been contemplated at all.

Neither the closest prior art alone or in combination with any of the other cited documents leads the skilled person to the presently claimed subject-matter.

Regarding this situation, the novel use of polyoxyalkylene sorbitan fatty acid esters for the prevention of clogging of filters in suspension culture processes is to be considered as involving an inventive step, i.e. the subject-matter of present claims 1-

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10 meets the requirements for the presence of inventive step.